A Single Institution-Based Retrospective Study of Surgically Treated Bronchioloalveolar Adenocarcinoma of the Lung

Clinicopathologic Analysis, Molecular Features, and Possible Pitfalls in Routine Practice

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Introduction: Prognostic evaluation of bronchioloalveolar carcinoma (BAC) from a homogenous population of Caucasian patients.
Methods: Retrospective analysis of resected BAC reclassified according to the 2004 World Health Organization classification of lung tumors. Analyzed variables are clinicoradiologic presentation, histologic subtypes, stage, epidermal growth factor receptor (EGFR) and HER2/neu immunohistochemical expression, *EGFR* exons 18, 19, and 21 mutations, *K-RAS* exon 2 mutation. Univariate and multivariate analyses of survival were performed.
Results: Of 40 patients analyzed, EGFR and HER2/neu expression were detected in 72% and 20%, respectively. HER2/neu expression significantly characterized mucinous BAC (46% versus 7%; $p = 0.014$). *EGFR* mutations were identified in 17% (30% in nonmucinous BAC and none in mucinous BAC; $p = 0.083$). *K-RAS* mutations were found in 42.5% (92% in mucinous BAC versus 18% in other types; $p < 0.0001$). Early stages (IA+IB) nonmucinous BAC had excellent prognosis: 5 years overall survival of 91% (100% for stage IA). Sixty six percent (4 of 6) of patients with multifocal disease died (two mucinous BAC and one nonmucinous BAC with recurrent disease). Seventy one percent (5 of 7) of patients with pneumonic-like tumor (all mucinous BAC) died of recurrent/progressive disease. Stage ($p = 0.004$) and histologic classifications ($p = 0.035$) resulted as independent prognostic factors at multivariate analysis.
Conclusions: Early stage nonmucinous BAC has excellent prognosis, whereas mucinous BAC presents a poor prognosis. Locally advanced nonmucinous BAC has a poor prognosis: the diagnosis of nonmucinous BAC in large tumors should be interpreted with caution given the possible presence of invasive areas in incompletely sampled tumor. Coexpression of EGFR and HER2/neu in mucinous BAC could be considered for future trials on target therapies even in Caucasian population.

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Bronchioloalveolar carcinoma (BAC) is considered a subtype of pulmonary adenocarcinoma presenting unique clinical and pathologic features. The latest 2004 World Health Organization (WHO) and the International Association for the Study of Lung Cancer (IASLC) classifications of lung tumors define BAC as an adenocarcinoma without stromal, pleural, or blood vessels infiltration.[1] These strict criteria have lead to a significant cultural change in the relationship between adenocarcinoma and BAC. Most of tumors diagnosed as BAC before the release of the current WHO classification were instead invasive adenocarcinomas with some percentage of BAC component at the tumor periphery and should now be correctly reclassified as mixed acinar adenocarcinoma with BAC features.[2] BAC, according to the current strict histologic diagnostic criteria, represents an exceedingly rare neoplasm if we consider that what was previously called BAC only accounted for 2 to 5% of all nonsmall cell lung carcinoma (NSCLC).[1,2] Therefore, it is incorrect to compare results of studies on BAC before and after the third edition of the classification of lung tumors. Recently, attempts have been made to classify small peripheral adenocarcinomas into subgroups according to the pattern of tumor growth,[3] and the presence of BAC features seems to predict prognosis in mixed adenocarcinoma.[3–6] Despite these recent evidences, the real prognosis of BAC tumors diagnosed according to the current 2004 WHO classification is still controversial, and in particular, the prognostic value of BAC subtypes (nonmucinous BAC versus mucinous BAC versus mixed subtype BAC) and the relationship among pathologic features, clinicoradiologic presentation, and response to surgical treatment are still unclear. Moreover, molecular evaluation of these tumors has revealed a significant difference between nonmucinous BAC and mucinous

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BAC which seem as two distinct entities, in particular, when K-RAS and epidermal growth factor receptor (EGFR) alterations are considered.[7] These genes are deeply involved in early molecular stages of pulmonary adenocarcinogenesis, show mutually exclusive mutation, and are significantly associated with BAC subtypes and patients clinicopathologic features.[7,8] Of note, clinicopathologic and genetic results on BAC mainly derive from studies on East Asian population, but it is suggested that several differences between Caucasian and Asian ethnicity may exist.[7,8]

The aim of this single-institution retrospective study is to investigate the prognostic effect of the current 2004 WHO histologic classification in surgically resected BAC tumors in a population of Caucasian patients. We also analyze the prognostic role of clinicopathologic features and genetic abnormalities of K-RAS and EGFR.

PATIENTS AND METHODS

Files from the archives of the Section of Pathologic Anatomy of the University of Modena and Reggio Emilia were retrospectively reviewed to retrieve all the patients with a diagnosis of BAC of the lung who underwent surgical resection at the Division of Thoracic Surgery of the University of Modena and Reggio Emilia between January 1995 and December 2007. All the slides of these selected patients were then reviewed by two pathologists of our group (G.R. and E.T.) and reclassified according to the diagnostic criteria set by the 2004 WHO classification of lung tumor,[1]in which BAC is defined as a "growth of neoplastic cells along preexisting alveolar structures (lepidic growth) without evidence of stromal, vascular, or pleural invasion." Three BAC subtypes were recognized as follows: mucinous, nonmucinous, and mixed types. All the cases analyzed consisted of surgical specimens that were routinely fixed in 10% buffered formalin. After gross examination, the sampled tissues were embedded in paraffin blocks. Complete sampling of tumor areas was performed in 34 of 40 cases, whereas six cases were incompletely examined because of the large size the tumor occupying almost the entire pulmonary lobe. A mean of four (range, 2–12) hematoxylin and eosin-stained slides per tumor was available. In doubtful cases of pleural invasion, histochemical stain for elastic fibers (Weigert and/or Van Gieson stains) was performed. All the patients were Caucasian and no East Asian cases were present in the study population.

For all patients, the following clinical and pathologic features were recorded: age at diagnosis, gender, smoking history (never smokers—lifetime exposure to $<$100 cigarettes; former smokers—patients quit smoking $>$3 years before diagnosis; and current active smokers), clinicoradiologic presentation at imaging evaluation (solitary, multifocal, or pneumonic), surgical procedure, postoperative mortality (within 30 days from the surgical procedure), pathologic staging, EGFR and HER2/neu immunohistochemical expression, *EGFR* and *K-RAS* genes mutations. All the patients were staged according to the 1997 International Association for the Study of Lung Cancer/Union Internationale Contre le Cancer Tumor, Node, Metastasis staging system.[9] Patients with an indeterminate tumor size because of a pneumonic-like presentation were staged as TX. Patients with multiple nodules of the same tumor histologic type in the same lobe were classified as IIIB, whereas multiple nodules in different lobes were considered as stage IV. Complete resection was defined as removal of the primary tumor and all accessible hilar and mediastinal lymph nodes with no residual neoplastic tissue left. Anatomic resection was considered as the standard procedure for all cases. Minor resections were reserved for patients with compromised pulmonary function. All the patients underwent mediastinal lymph node dissection. No patient underwent adjuvant therapy with EGFR tyrosine kinase inhibitors.

Clinical and follow-up data were collected from pathologic reports, clinical charts, referring physicians, or directly from the patients' families. This is a retrospective descriptive study, and all information was collected and used to perform anonymous and aggregate statistical analysis. The institutional review board of Policlinico of Modena has approved this study.

Immunohistochemical Analysis

The antibodies used in the study and their technical characteristics are the following: EGFR (prediluted; protease pretreatment; clone 31G7; Ventana, Tucson, AZ) and HER2/neu (1:150 dilution; microwave; clone CB11; Novocastra, Newcastle upon Tyne, UK). Negative and positive controls were included in each batch. For each antibody, the percentage of positive cells and the intensity of staining (0: negative; 1+: weak; 2+: moderate; and 3+: strong) were recorded. A tumor was considered positive when at least 20% of the neoplastic cells reacted with a 2+ or higher intensity on the relevant subcellular localization (cytoplasmic and/or membranous).

DNA Extraction and Sequencing Analysis

Molecular analysis of *EGFR* exons 18, 19, and 21 and *K-RAS* exon 2 was performed by direct sequencing polymerase chain reaction as described in a previous study by our group[10] and in both forward and reverse directions.

Statistical Analysis

The descriptive analysis was expressed in terms of frequency, median and SD. Frequencies were compared with the χ^2 test, for categorical variables; Fisher exact test was used for small samples. *t* test and analysis of variance were performed when comparing continuous variables. The overall and disease-free survivals were calculated according to the Kaplan-Meier method. Deaths from noncancer-related causes were considered as withdrawals. Date of death or recurrence represented the end points of follow-up for each type of survival analysis. Univariate analysis of overall and disease-free survivals was performed using the log-rank test. Variables resulting statistically significant at univariate analysis were evaluated in a multivariate analysis using Cox regression models. A probability value of less than 0.05 was considered statistically significant.

RESULTS

Of 1468 patients with a diagnosis of NSCLC who underwent surgical resection with curative intent in the ana-

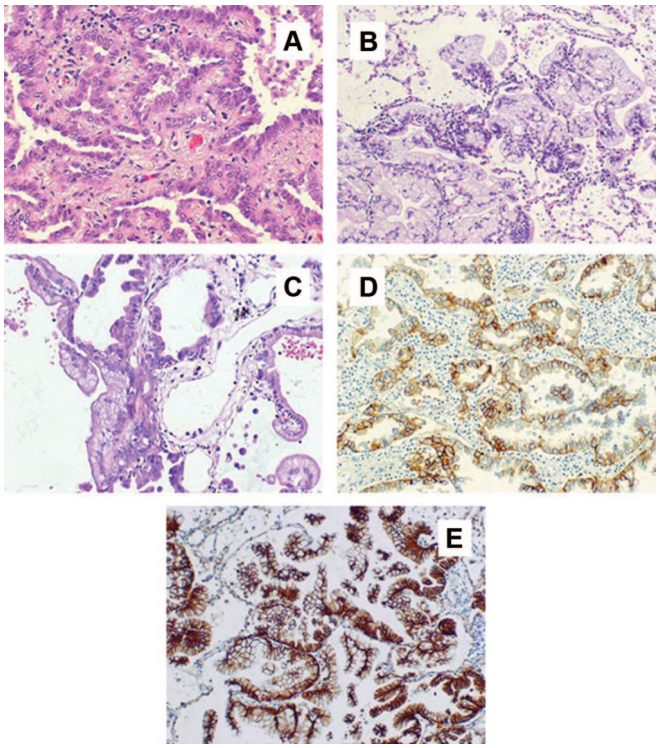

FIGURE 1. Example of a nonmucinous BAC (*A*, hematoxylin and eosin; original magnification, ×155), mucinous BAC (*B*, hematoxylin and eosin; original magnification, ×100), mixed BAC (*C*, hematoxylin and eosin; original magnification, ×155). Positive immunostaining for EGFR in a nonmucinous BAC (*D*, EGFR, immunohistochemistry, magnification ×155), and HER2/neu in a mucinous BAC (*E*, HER2/neu, immunohistochemistry, magnification ×100).

TABLE 1. Patients Characteristics

Variable	No. of Patients (%)
Gender	
Male	23 (57)
Female	17 (43)
Smoking history	
Current	29 (73)
Former	4 (10)
Never	7 (17)
Clinical presentation	
Solitary	27 (66)
Multifocal	6 (15)
Pneumonic	7 (19)
Surgical procedure	
Lobectomy	30 (75)
Pneumonectomy	1 (2)
Wedge resection	9 (23)
pStage	
IA	12 (30)
IB	12 (30)
IIA	1 (2)
IIB	1 (2)
IIIA	1 (2)
IIIB	2 (5)
IV	4 (10)
TX	7 (19)
Histological classification	
Nonmucinous	20 (50)
Mucinous	13 (32)
Mixed	7 (18)
EGFR expression	
Positive	29 (72)
Negative	11 (28)
HER2/neu expression	
Positive	8 (20)
Negative	32 (80)
EGFR mutation	
No mutation	33 (83)
Exon 18	0
Exon 19	1 (2)
Exon 21	6 (15)
RAS mutation	
No mutation	23 (57.5)
Exon 2	17 (42.5)

lyzed period, a total of 127 cases (9%) were initially collected. After histologic review at multiheaded microscope, 87 cases resulted as mixed acinar invasive adenocarcinoma with BAC features and were not included in the statistical analysis (pleural invasion and stromal invasion was present in 16 and 71 cases, respectively). Forty patients (3% of all NSCLC resected in the analyzed period) with a BAC diagnosis complying with current diagnostic criteria were finally evaluated (Figures 1*A–C*). Clinical, pathologic, and genetic features of the patients are listed in Table 1. Mean age was 65 years (median 66, SD 7.91, range 37–80 years). No postoperative death was reported.

Significant correlations between histologic classification, pathologic stage, clinical presentation, and genetic abnormalities were recorded and summarized in Table 2. Nonmucinous BAC presents more frequently in a solitary form ($p = 0.012$) at an early stage ($p = 0.004$), whereas mucinous BAC is more frequently associated to a multifocal disease or a pneumonic-like clinical pattern ($p = 0.012$) in more advanced stages ($p = 0.004$). Overall, EGFR and HER2/neu expression were noted in 72% and 20%, respectively. However, EGFR expression did not seem to be significantly associated to BAC subtypes (Figure 1*D*), whereas HER2/neu expression significantly characterized mucinous BAC (46% versus 7% in the other types; $p = 0.014$; Figure 1*E*). Overall, *EGFR* mutations were identified in 17% (six exon 21 missense and one exon 19 deletion mutations), whereas *K-RAS* mutations were found in 42% of BAC (eight G12C; five G12V; three G12D; one G12A). As reported in Table 2, the mutation profile was significantly different in the histologic subtypes. *EGFR* mutations were also strongly related to never smoking status (57% versus 9%; $p = 0.002$) and in female gender (29% versus 8%, $p = 0.05$). No significant correlation was found among *K-RAS* mutation, gender, and smoking habit.

TABLE 2. Correlation Between 2004 WHO Histological Classification and Clinical Presentation, Pathological Stage, and Molecular Setup for *EGFR* and *KRAS* Mutations

	Histologic Subtypes, No. of Patients (%)			
Variable	**Mucinous**	**Nonmucinous**	**Mixed**	***p***
Clinical presentation				0.012
Solitary	5 (38)	17 (85)	5 (71)	
Multifocal	2 (16)	2 (10)	2 (29)	
Pneumonic	6 (46)	1 (5)		
pStage				0.004
IA	1 (8)	10 (50)	1 (14)	
IB	4 (30)	5 (25)	3 (43)	
IIA+IIB+IIIA		2 (10)	1 (14)	
IIIB			2 (29)	
IV	2 (16)	2 (10)		
TX	6 (46)	1 (5)		
EGFR mutation				0.083
No mutation	13 (100)	14 (70)	6 (86)	
Exon 18				
Exon 19		1 (5)		
Exon 21		5 (25)	1 (14)	
RAS mutation				0.001
No mutation	1 (7)	17 (85)	5 (71)	
Exon 2	12 (93)	3 (15)	2 (29)	

Analysis of Survival

Follow-up was completed for all patients by December 2008 (median 48 months, range from 2 to 132 months). For all patients, the overall 5 years survival rate was 64%, with a median survival time of 69 months. Five years disease-free survival was 51% with a median disease-free survival time of 28 months.

Nineteen patients (48%) developed tumor recurrences/metastases after pulmonary resection: pulmonary recurrences/metastases in 14 patients (35%) and metastases in extrathoracic sites in five patients (12%: three brain, one bone, and one liver metastases). All the patients who developed extrapulmonary metastases resulted incompletely sampled at gross examination. Ten patients (25%: four nonmucinous BAC, four mucinous BAC, and two mixed BAC) with solitary intrathoracic recurrence underwent a second surgical resection. Five years overall survival of reoperated patients was 60% (four patients died of a second recurrence: three mucinous BAC and one mixed BAC).

Overall, 18 patients died during the follow-up period: four patients (all with nonmucinous BAC) without disease recurrence died because of lung noncancer-related causes, and 14 patients (eight mucinous BAC, three nonmucinous BAC, and three mixed BAC) died of recurrent-progressive disease. Five patients with recurrent disease (three nonmucinous BAC, one mucinous BAC, and one mixed BAC) were alive at the end of the follow-up.

At univariate analysis of survival, clinicoradiologic presentation, pathologic stage, and histologic pattern significantly affected overall and disease-free survivals (Figures 2*A–D*). The 5 years overall and disease-free survival for solitary lesions were 73% and 63%, for multifocal lesions 50% and 25%, and for pneumonic-like tumors 28% and 28%, respectively ($p = 0.048$ for overall survival and $p = 0.051$ for disease-free survival). The 5 years overall and disease-free survival for mucinous BAC were 44% and 35%, for nonmucinous BAC 81% and 67%, and for mixed BAC 47% and 28%, respectively ($p = 0.021$ for overall survival and $p = 0.016$ for disease-free survival). The 5 years overall and disease-free survival were 79% and 69%, respectively for stage IA+IB, 34% and 28% for advanced stages ($p = 0.006$ for overall survival and $p = 0.020$ for disease-free survival).

Other clinical (age, gender, type of surgery, and smoking habit), immunohistochemical (EGFR and HER2/neu expression), and genetic (*EGFR* and *K-RAS* mutations) variables were not significantly related to survival at univariate analysis. However, a trend toward better overall survival was recorded for patients with *EGFR* mutations (5 years overall survival of 80% for *EGFR* mutation and 56% for absence of *EGFR* mutation, $p = 0.064$) and for patients without *K-RAS* mutation (5 years overall survival of 52% for absence of *K-RAS* mutation and 76% for *K-RAS* mutation, $p = 0.065$).

Within solitary forms, 5 years overall survival for stages IA and IB was 91% and 69%, respectively. No survival analysis could be computed for solitary stages II and IIIA because of a limited number of cases (three patients). Survival analysis according to histologic patterns in solitary forms at early stages (IA and IB) revealed a significantly increased overall and disease-free survivals for nonmucinous BAC compared with mucinous BAC and mixed BAC (Figures 2*G*, *H*). In solitary early stage (stages IA and IB), the 5 years overall survival and disease-free survival were 91% and 74% respectively for nonmucinous BAC, and 66% and 45% for mucinous BAC/mixed BAC ($p = 0.044$ for overall survival and $p = 0.042$ for disease-free survival). In particular, the 10 patients with stage IA solitary nonmucinous BAC were all alive at the end of follow-up (5 years overall survival of 100%).

The small subgroup of patients with multifocal disease and pneumonic-like disease (six and seven, respectively) did not allow a reliable analysis of survival. Four of six patients (66%) with multifocal tumor died (two mucinous BAC and one nonmucinous BAC for recurrent disease and one nonmucinous BAC for noncancer-related cause). Two patients with multifocal mixed BAC were alive at 37 and 47 months after surgery, one with a recurrent disease. Five of seven patients (71%) with pneumonic-like BAC died of recurrent pulmonary disease. All these patients had mucinous BAC. One patient with mucinous pneumonic-like BAC was alive without disease at 72 months and one patient with nonmucinous pneumonic-like BAC was alive with recurrent disease at 77 months.

All the clinical and pathologic variables that resulted significantly related to survival at univariate analysis were examined in a multivariate analysis (Table 3). Stage ($p = 0.004$) and histologic classifications ($p = 0.035$) were confirmed as independent prognostic factors affecting overall survival, whereas no variable resulted significantly related to disease-free survival.

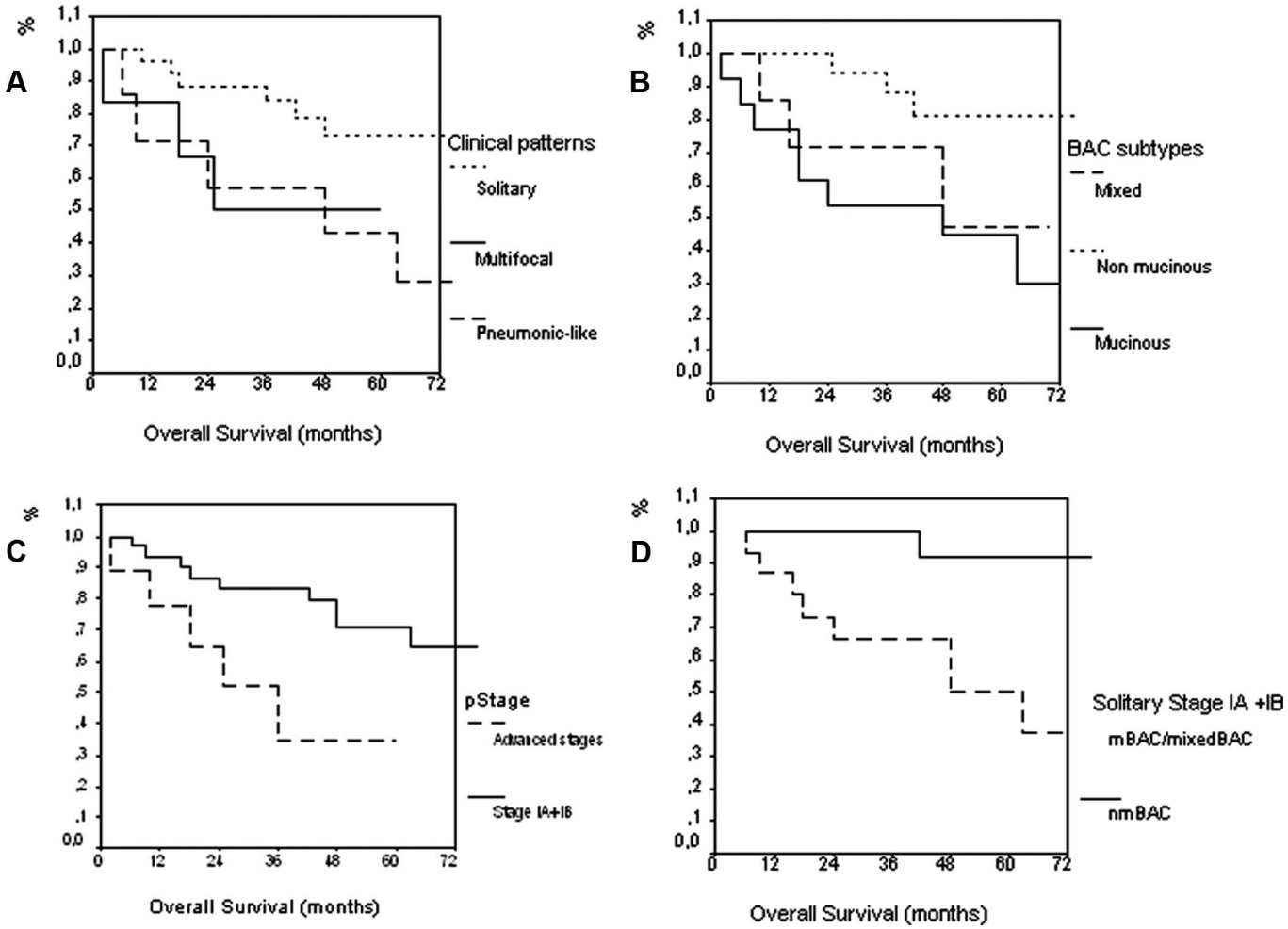

FIGURE 2. Kaplan-Meier survival curves. *A*, Overall survival for clinical pattern ($p = 0.048$); *B*, Overall survival for BAC histologic subtype ($p = 0.021$); *C*, Overall survival for pathologic stage ($p = 0.006$); *D*, Solitary early stages (stages IA and IB; $p = 0.044$).

DISCUSSION

The definition of pulmonary adenocarcinoma and the concept of BAC in particular are still matter of debate and investigation among surgeons, radiologists, clinicians, and pathologists too,[1,2] namely for the following reasons: (a) the epidemiologic changes of lung cancer incidence with adenocarcinomas greatly overriding other histotypes[11]; (b) the discovery of somatic *EGFR* and *K-RAS* mutations in adenocarcinomas with significant impact on treatment with targeted therapies against EGFR[12,13]; (c) the new histologic definition of BAC as a noninvasive lung cancer, with the possible intrinsic curative significance once completely resected[1,3]; and (d) the emerging clinicopathologic, molecular, and possibly therapeutical models of lung adenocarcinogenesis according to patient's gender and smoking status.[12–14] The results of our study confirm that even in the Caucasian population the current concept of BAC involves at least two distinct and unrelated tumor entities with peculiar clinicoradiologic presentation and molecular and prognostic features. The nonmucinous BAC subtype usually presents as a solitary lesion or more rarely with multiple lesions or pneumonic-like expression. The prognosis for this subtype of BAC tumor is usually favorable even though our survival results are lower than those previously reported by Japanese authors.[3–5] This discrepancy can be explained by the different selection criteria and morphologic classification adopted. Our retrospective study includes a relatively large amount of advanced stage cases, whereas the surgical series by Japanese authors usually derive from computed tomography screening programs where only representing early stages of the disease. In our experience, a significant percentage of locally advanced nonmucinous BAC presented extrapulmonary metastases. As suggested in the 2004 WHO classification, the diagnosis of nonmucinous BAC should not only be based on surgical specimens but also the tumor should be completely sampled to avoid invasion. In our experience, six tumors of large size were not entirely sampled. The retrospective design of this study did not allow evaluation of further blocks of these incompletely sampled neoplasms. In these cases, the available blocks showed a BAC tumor, but the presence of

TABLE 3. Multivariate Cox Survival Analysis According to Stage, Histologic Classification, and Clinical Presentation

	Overall Survival		Disease Free Survival	
Variable	**HR (CI 95%)**	***p***	**HR (CI 95%)**	***p***
Clinical presentation				
Solitary vs. multifocal/pneumonic	0.205 (0.033–1.251)	0.086	0.652 (0.050–8.369)	0.249
Pathologic stage				
IA and IB vs. others	0.066 (0.010–0.429)	0.004	0.685 (0.191–2.458)	0.416
Histologic classification				
nmBAC vs. mBAC/mixedBAC	0.223 (0.055–0905)	0.035	0.403 (0.046–3.353)	0.542

HR, hazard ratio.

invasion cannot be reasonably excluded. Five of six patients experienced extrapulmonary metastases. This finding raises the issue on the importance of gross examination in diagnosing BAC. Moreover, Japanese authors adopted the Noguchi's classification system,[3] in which type A and B peripheral (replacing type) adenocarcinomas showed an excellent prognosis after surgical resection. The prognosis of early stages nonmucinous BAC of our surgical series reflects these results, with a 5-year overall survival of 100% for stage IA and 91% for stage IB because of one nonlung cancer-related death. In our surgical series, no patient with an early stage nonmucinous BAC died of a cancer-related cause. Early stages nonmucinous BAC diagnosed according the 2004 WHO criteria have the same excellent prognosis of types A and B Noguchi's peripheral adenocarcinoma. Based on these promising results, some authors advocate limited resection without mediastinal lymph nodes dissection for these tumors when they are more than 2 cm of diameter.[15–17] In our series, only nine patients with compromised pulmonary function underwent limited resections. Although, at univariate analysis of survival, the prognosis of these patients did not significantly differ from that of patients treated with major resection, definitive conclusions on the ideal type of resection cannot be drawn from the current small series of BAC. Until the results of ongoing Japanese trials on limited resection for peripheral lesions are available, major pulmonary resections with mediastinal lymph nodes dissection should be considered the gold standard for these lesions.

Recent studies have demonstrated no survival differences after surgery between solitary and multifocal nonmucinous BAC.[18] Although only two cases (5%) of multifocal nonmucinous BAC are present in our series, we suggest that such tumors should be surgically treated whenever possible because satisfactory results can be obtained when BAC has a nonmucinous component. In general, BAC presents pulmonary recurrences (usually ipsilateral) more frequently than other types of NSCLC.[17] The optimal management of recurrent disease is still debated. In our series, 25% of patients experienced a pulmonary recurrence and was reoperated with good survival results. In such cases, limited resections should be preferred considering the high rate of expected pulmonary recurrences that should be subsequently amenable of reoperation.

Mucinous and mixed BAC usually present as pneumonic-like masses. Patients with pneumonic-like BAC generally present a poor prognosis after surgery,[19] with a high percentage of patients early developing pulmonary recurrences and/or, more rarely, distant metastases. Moreover, recurrences are usually not amenable of reoperation. In our series, 71% of patients showing a pneumonic-like mucinous BAC died of respiratory failure because of widespread pulmonary involvement, therefore supporting the idea that tumor cell aerogenous spread throughout the alveoli should be considered as an invasive factor similar to what happens in pleura, stroma, or blood vessels. Basically, mucinous BAC could remain a variant of an invasive form of adenocarcinoma according to the current classification of lung cancer. This issue supports the diagnostic criteria proposed in the new classification of lung adenocarcinoma recently presented at the 13th World Congress of Lung Cancer.[20]

The hypothesis that mucinous BAC and nonmucinous BAC are two clear-cut distinct entities is strongly supported by their entirely different immunoprofile, obtained using a panel of diagnostic markers (TTF-1, cytokeratins 7 and 20, MUC2 and MUC5, and surfactant protein),[21] and their quite peculiar molecular setup for *EGFR* and *K-RAS* genes mutations, indicating two different pathways of oncogenesis.[8,22–25] As confirmed by a sound body of evidence,[7,8,22,23,26] also in our study *EGFR* mutations were significantly identified in nonmucinous BAC (30%), whereas *K-RAS* mutations characterized mucinous BAC (92%). As expected, even nonsmoking status and female gender were strongly related to *EGFR* mutations, whereas tumors in smoking men had a significant association with *K-RAS* mutations.[7,8] The high ratio of *EGFR* exon 21/exon 19 mutations detected in our study could somehow appear in discrepancy with that commonly reported in similar studies. However, an equal distribution of *EGFR* mutations on exons 19 and 21 has been reported in a large series of NSCLC by Marchetti et al,[24] whereas Hsieh et al.[25] found *EGFR* exon 21 and exon 19 mutations in 76% and 17%, respectively, analyzing a series of 35 adenocarcinomas. In addition, our study reported an interesting and significant association between HER2/neu expression and mucinous BAC. Although genetic profiles have not revealed a significant prognostic impact in our series, all these observations basically point out that the concept of

BAC currently includes at least two different oncogenetic pathways. These results could be considered in future trials aimed at evaluating specific target therapies for different histologic subtypes of BAC.

Given the unavoidable limitations because of the retrospective design of our study, definitive conclusions cannot be drawn. However, some key messages emerged from our results. BAC diagnosed according to the current WHO classification are rare lung tumors in the routine practice, accounting for approximately 2% of all resected NSCLC, and their histologic subtypes represent independent prognostic factors. The definition of BAC, when appropriately applied on histologic analysis, includes different entities with different clinical, pathologic, and genetic features possibly leading to different prognosis and therapeutic approaches. As previously reported in several works from East Asia, early stage nonmucinous BAC can be cured with surgical resection alone in almost all cases, presenting an acceptable prognosis even when pulmonary recurrences are surgically treated. However, we point out that the diagnosis of BAC requires the evaluation of the entire surgical specimen. This issue can present some pitfalls in the diagnosis of large size tumors, which ultimately lead to unexpected poor survival results in locally advanced nonmucinous BAC. Mucinous BAC presents a poor prognosis after surgical resection because of the high local recurrence rate leading to respiratory failure. The aerogenous spread along the alveoli characterizing mucinous BAC (or "mucinous adenocarcinoma with lepidic pattern," as proposed by the new IASLC/American Thoracic Society/European Respiratory Society classification of lung adenocarcinomas) should be considered as a morphologic component of invasive parameters. Given the close association between *K-RAS* mutations and mucinous BAC, and the significant combined expression of EGFR and HER2/neu in this subtype, an alternative approach using humanized monoclonal antibodies against EGFR and/or HER2/neu could be evaluated in future trials including mucinous BAC.

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